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Introduction

The purpose of this grant project is to identify and characterize methods for ablation of CML stem cells. Numerous studies have shown that malignant stem cells lie at the heart of CML disease; therefore, specific targeting of these cells is essential for more durable remission and cure. To this end, we are employing both mouse and human model systems to study the nature of CML stem cells and to identify effective drug combinations that will preferentially destroy malignant stem cells while sparing normal cells.

Body

PLEASE NOTE: Although this report covers the first year of the grant project, it should be noted that research activities were only conducted from the beginning of the grant period on April 15th, 2003 until August 31st, 2003. At that time Dr. Jordan moved his laboratory from the University of Kentucky to the University of Rochester. Since that time, Dr. Jordan has been in the process of completing the requirements for transfer of the grant. The University of Kentucky only released a final financial report in February 2004 and still has not completed transfer of unexpended funds from year one. Hence, laboratory activities have been suspended pending completion of the grant transfer. This report consequently documents progress for the 4.5 month period prior to August 31st.

SOW task #1: To investigate apoptosis induction for human CML stem cells.

Studies to investigate the effects of proteasome inhibitors and idarubicin on CML stem cells were initiated. Preliminary data indicate a statistically significant reduction in CML CD34+ cells in comparison to normal controls. Further studies are pending upon transfer of the grant to the University of Rochester. In addition, we have collected 8 CML specimens and isolated DNA to analyze for the presence/function of p53.

SOW task #2: To analyze the role of NF-kB in the biology of CML stem cells.

These studies are not scheduled to begin until month 12 of the project

SOW task #3: To investigate the in vivo biology of CML stem cells in response to apoptotic stimuli.

Experiments to characterize the response of CML cells to PS341 were initiated. Studies to date have examined how cells transformed with the BCR/ABL oncogene are affected. The figures below demonstrate that PS341 is preferentially cytotoxic to BCR/ABL transformed cells in comparison to normal controls.

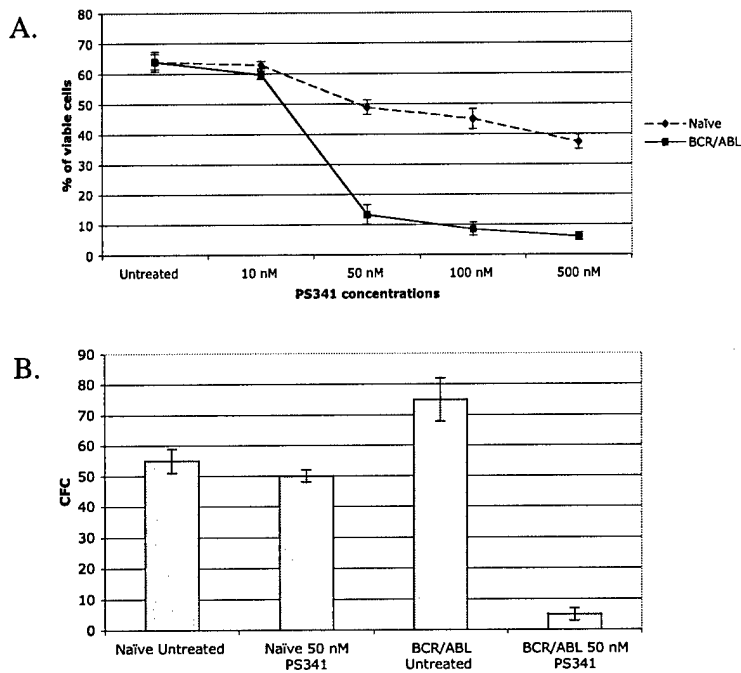


Figure 1: Effect of PS-341 on the growth of BCR/ABL transduced cells. a. Annexin V labeling of naïve or BCR/ABL-transduced cells treated with different concentrations of PS341 for 18 hrs *in vitro*. b. *In vitro* Colony-forming Cell (CFC) assays of untreated or PS341 (50 nM) treated marrow cells after 18 hrs of culture. Colony formation was evaluated after 10 days.

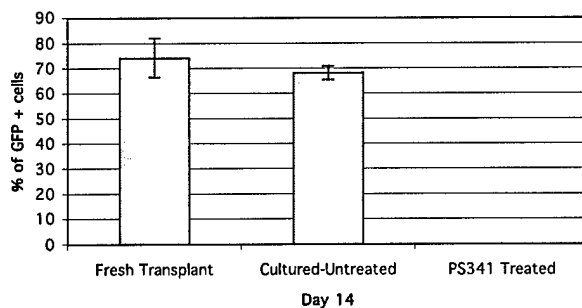


Figure 2: Effect of PS-341 treatment on transplantation of cells in recipient mice. Engraftment of BCR/ABL transformed cells was determined after 18 hrs of culture \pm 50 nM PS341 as compared to freshly transplanted BCR/ABL transformed cells (four animals were analyzed from each group). Flow cytometric analysis of peripheral blood from each group was performed 14 days following transplantation into sublethally (600 rad) irradiated recipients. The percentage of GFP+ cells indicates the level of leukemic cell engraftment.

Key Research Accomplishments

Established preferential toxicity of PS-341 for BCR/ABL transformed cells.

Reportable Outcomes

Manuscript in preparation that describes our studies of PS-341 for BCR/ABL transformed cells.

Conclusions

The initial findings suggest that PS-341 may be effective for treatment of chronic phase CML.

References

N/A

Appendices

N/A